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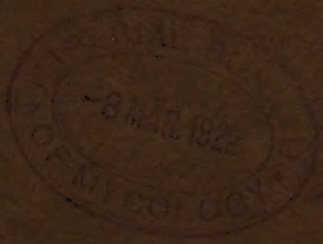

OBSERVATIONS ON A FUNGUS ENEMY OF THE WALNUT APHIS IN SOUTHERN CALIFORNIA

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OBSERVATIONS ON A FUNGUS ENEMY OF THE WALNUT APHIS IN SOUTHERN CALIFORNIA[1]

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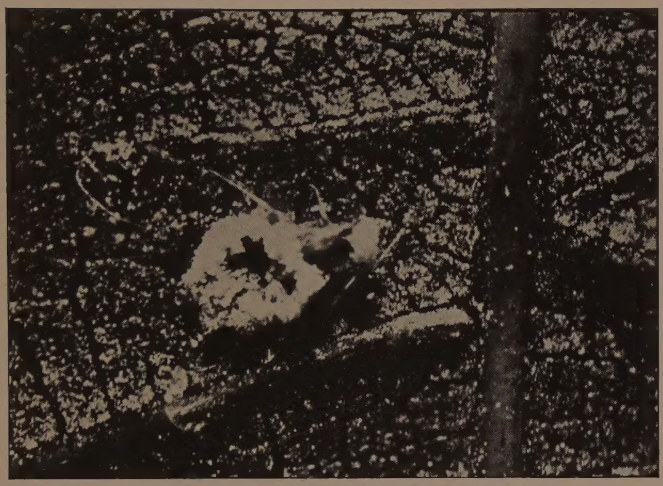
Introduction

In the spring and early summer of 1917 the walnut aphis (*Chromaphis juglandicola* Kalt.) was very abundant in the various walnut growing sections of Southern California. In many places the infestations were so severe that control measures were inaugurated. This was especially true in the vicinities of Santa Barbara, Santa Paula, El Monte and Tustin. Several growers dusted their trees with combinations of dry sulfur and tobacco dust, in an attempt to control both the aphids and the walnut blight. Under the direction of the Department of Entomology of the Citrus Experiment Station, portions of the walnut groves of the San Joaquin Fruit Company, near Irvine, Orange County, were sprayed in the latter part of May, with nicotine sulfate and lime sulfur, and with nicotine sulfate and soap, with excellent results in controlling the aphids. A grove near Santa Paula was sprayed about the same time with nicotine sulfate and soap under the direction of the Ventura County Horticultural Commissioner, with equally good results.

As stated above the infestation of walnut aphis was exceedingly heavy, and gave promise of causing considerable loss. Even in Riverside County where ordinarily this aphis is of no importance whatever, a severe infestation occurred during the latter part of May and in June. One hundred leaves from a tree in Riverside on June 8, showed an average of about 70 aphids per leaf, which is an extremely large infestation for that vicinity. On June 6, in a grove in Santa Ana, an average of 106 aphids per leaf was counted, while the grower informed the writers that two weeks earlier there had been many more present. In the Thorpe grove near Santa Paula on June 11, there was found to be an average of 62.5 aphids per leaf, and on the Limoneira Rancho a few days later, about 22 per leaf were noted. The great abundance of aphids was also observed in Santa Barbara County at Carpenteria, Naples and Santa Barbara; in Los Angeles County at El Monte, Spadra and Walnut; and in Orange County at Anaheim, Orange and Tustin.

[1] Paper No. 49, University of California, Graduate School of Tropical Agriculture and Citrus Experiment Station, Riverside, California.



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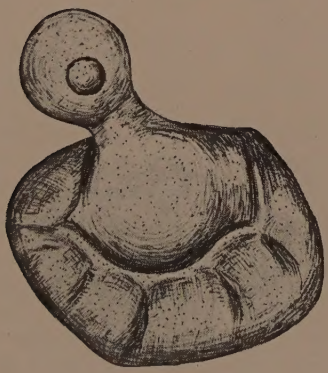

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Field Investigations

Several groves in the vicinity of El Monte were heavily infested and many of them were treated. The observations were confined to the groves of the Cogswell Ranch which were dusted with various combinations of sulfur and tobacco dust during the latter part of May. A few days later the owner noticed that the infestation was considerably lighter. On June 4, the writers examined these groves and found that the aphids to a large extent had disappeared. Bodies of many dead aphids were observed still clinging to the leaves, and these were thought at first to have been killed by the dust, but they were also observed on untreated trees. It was further noticed that live aphids were no more numerous on the untreated than on treated trees. A microscopical examination of the bodies of the dead aphids brought out the fact that they were infected with a fungus which later proved to be a new species of *Entomophthora.*

The discovery of aphids infected with this fungus resulted in a more thorough examination of the conditions on June 9. Table 1 gives the results of counts made on the various groves.

Table 1—Walnut Aphis in Cogswell Groves, El Monte, July 9, 1917

	Number of leaves	Number of aphids per leaf	Percentage of aphids infected	Number of living aphids per leaf
A Treated	30	40.07±2.87	87.02±1.51	5.93±0.65
B Treated	50	17.56±0.94	73.12±2.27	4.72±0.64
C Treated	50	28.94±1.50	71.73±1.73	8.18±0.69
D Untreated	49	42.18±2.80	88.56±1.17	5.16±0.39
E Untreated	40	38.60±2.67	83.81±1.33	6.17±0.94

An examination of this table shows that the average number of aphids per leaf ranged from 17 to 42 in the various groves, but on account of this fungus the average of living aphids was only 5 to 8 per leaf, the percentage of infection being about the same in all groves, both treated and untreated. It must be borne in mind that the number of aphids per leaf does not represent a true record of the previous infestation, for many of the dead aphids had either fallen to the ground or been blown off the leaves. However, this table does show that the fungus was a very important factor in the elimination of the aphids.

Observations were made in Ventura County about this time, where it was found that aphids were present in great numbers but that the fungus was also present. On May 11 one grove near Santa Paula was found to be heavily infested. It was also observed that a small percentage of infected aphids were present. On June 11 not a single

living aphid could be found in this grove, although the mummified bodies of dead aphids were abundant. Unfortunately no counts were made at either examination. It is not claimed by the writers that the fungus was the sole agent in the eradication of the aphids on this grove, for large numbers of ladybird beetles and larvæ, and of syrphid fly larvæ were observed at the time of the first examination. However, the fact that such a large number of infected bodies was present, when living aphids were absent, proves that the fungus was an important agency in controlling the aphis.

Natural Factors in the Control of the Walnut Aphis

During the week June 11 to June 18, extremely high temperature and correspondingly low humidity prevailed throughout Southern California. In fact, that week was the hottest period for the time of year ever reported in Southern California. Table 2 gives the maximum temperatures recorded at various points for the week.

Table 2—Maximum Temperatures (F.°), Week Ending June 19, 1917[1]

June	12	13	14	15	17	18
Los Angeles	79	80	100	99	105	85
Santa Ana	79	87	104	95	97	83
Pomona	93	99	110	112	117	100
Riverside	98	105	112	114	117	103
Santa Barbara	76	80	92	105	115	77

This extreme heat resulted in the death of a large percentage of aphids throughout the walnut growing sections of Southern California, and was by far the most efficient check of the season. In only one case were any living aphids observed by the writers during the following week. This was on the Limoneira Rancho, Santa Paula, where on June 19 there was an occasional aphid found. Such aphids were found only on leaves well in the interior of large trees, where they were protected considerably from the heat.

Among the natural agents which contribute to the control of this aphid, the heat of summer can be mentioned as very important in certain sections. To this heat is largely due the credit for the practical absence of the walnut aphis in such interior sections as Riverside, Elsinore and Hemet. In harmony with this is the fact that in cool, humid weather the aphids seem to thrive best. The early part of the season of 1917 was cool and humid, and this condition lasted until June.

[1] These records were obtained from the Daily Weather Map, United States Department of Agriculture, Weather Bureau, Los Angeles, California, June 13–19, 1917, with the exception of those for Santa Ana which were obtained from the records of a thermograph placed in the Rohrer Grove, by the Department of Plant Pathology of the Citrus Experiment Station.

Insect Enemies

Insect enemies of the walnut aphis, particularly the predaceous enemies, are a very important factor in their control. Observations by the writers showed that these were present everywhere, in some cases being very abundant and apparently accomplishing a great reduction in the aphid infestations. Among the more common enemies may be mentioned the ashy gray ladybird beetle (*Olla abdominalis* Say). About the middle of May the eggs of this species were abundant in the vicinity of Santa Paula, El Monte, Walnut and Tustin. By the latter part of the month, the larvæ were present and feeding extensively on the aphids. These pupated in early June and the adults appeared shortly before the extreme heat. On the San Joaquin Fruit Company's groves, these were very abundant.

Another common predaceous enemy was *Hippodamia convergens* Guerin, the larvæ and adults of which were present from May to September in all groves examined by the writers. The third in abundance was the green lace wing (*Chrysopa californica* Coquillet), which was very plentiful in May and June in Ventura, Los Angeles and Orange Counties. From time to time larvæ of various syrphid flies, particularly *Catabomba pyrastri* (Linnæus), were noted, although never in any great numbers. Occasionally larvæ of *Sympherobius angustus* (Banks) were found, and in the vicinity of Walnut and Spadra, Los Angeles County, adults and larvæ of *Scymnus* sp. were observed. The inclination is to give predaceous enemies credit for any reduction of aphids not proven to be directly attributed to other agencies, as for instance the heat and fungus parasites.

Reinfestation of Groves

About six weeks after the hot weather mentioned above, an examination of the walnut groves of the San Joaquin Fruit Company was made. The aphids were still very scarce. A count of 186 leaves gave an average of 6.54 ± 0.30 aphids per leaf, showing that as yet they had not been able to reëstablish themselves to any great extent. By September 12, three months after the excessive heat, a count was again made, 50 leaves showing an average of but 12.02 ± 1.39 aphids per leaf. In that vicinity, however, the summer had been dry and warm, which together with the predaceous enemies had kept the infestation down. Table 3 gives the results of a count made on the Cogswell groves, El Monte, on September 12.

TABLE 3—WALNUT APHIS IN COGSWELL GROVES, EL MONTE, SEPTEMBER 12, 1917

	Number of leaves	Number of aphids per leaf	Percentage of aphids infected	Number of living aphids per leaf
B	30	84.57± 5.14	1.42±0.27	83.33± 5.14
D	10	63.10±12.71	1.43±0.96	62.20±12.81
F	20	51.30± 6.11	3.51±1.46	49.50± 6.17

An examination of this table shows that practically three months after the hot period, the aphids had been able to reinfest these groves quite extensively. At that time, however, the fungus, although present, was a negligible factor in control, since less than 2 per cent of the aphids were infected. The practical absence of the fungus was due undoubtedly to the summer heat and drought. However, at that season of the year, aphid infestations are of no commercial importance.

OBSERVATIONS AT SANTA ANA

In Santa Ana, Orange County, the Department of Plant Pathology of the Citrus Experiment Station had two walnut trees enclosed in a frame covered with cheesecloth for the purpose of studying walnut blight. Temperature and evaporation records were kept throughout the summer both within one of these frames and outside in the grove. It may be noted that the temperatures from August 1 to September 20 (the period of observations on the aphids) were practically the same outside as inside the frame, being but one or two degrees lower on the average inside. However, the humidity as obtained by the amount of evaporation recorded by the Livingston atmometers was higher within the tent than without. In fact, there was marked difference (see chart I).

It was noted (see Table 2) that in this locality the maximum summer temperature was 104 degrees F, which was considerably lower than in most of the sections of Southern California where any observations were made. Also the humidity there is comparatively high. As a result the infestation was not so depleted in June as in other localities. On July 26 under tent 1, a count of the aphids on 35 leaves showed an average of 44.51±4.09 aphids per leaf, with a mortality of 94.43±0.25 per cent caused by the fungus. On August 1 no live aphids could be found on this tree.

About every two weeks, from August 1 until September 20, counts were made of the number of aphids on leaves from the tree under tent 2, and on trees in the surrounding grove. The results of the counts are included in Tables 4 and 5.

CHART I
RELATIVE EVAPORATION

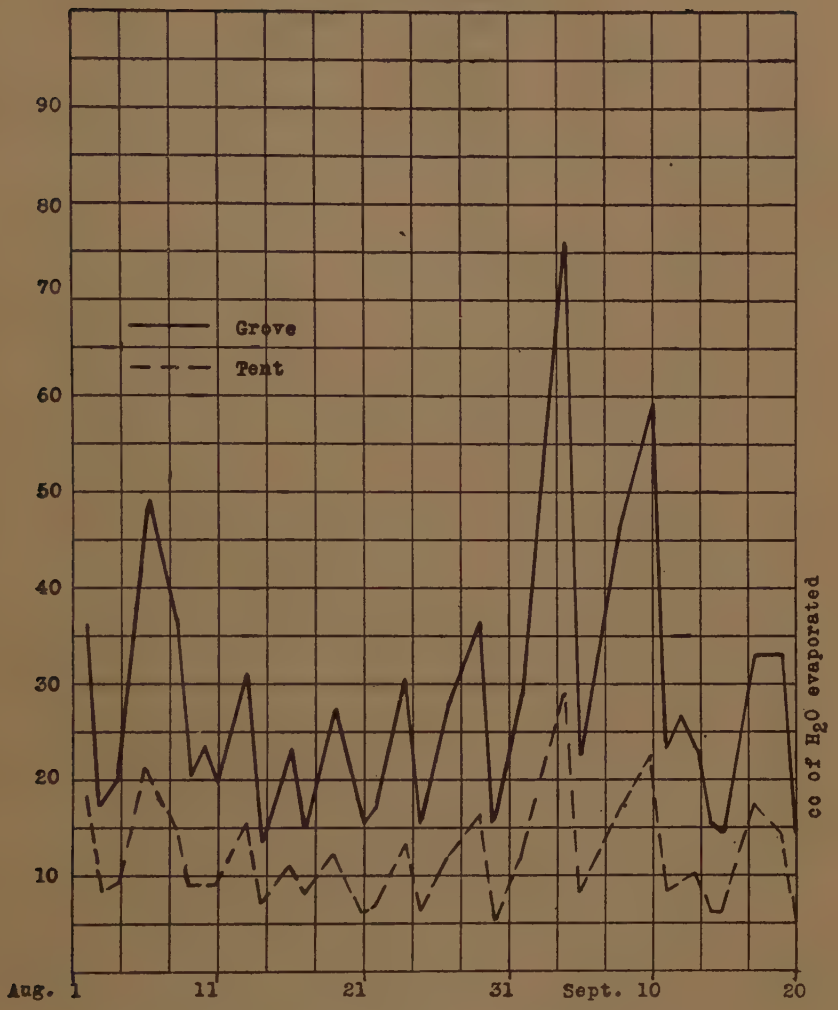


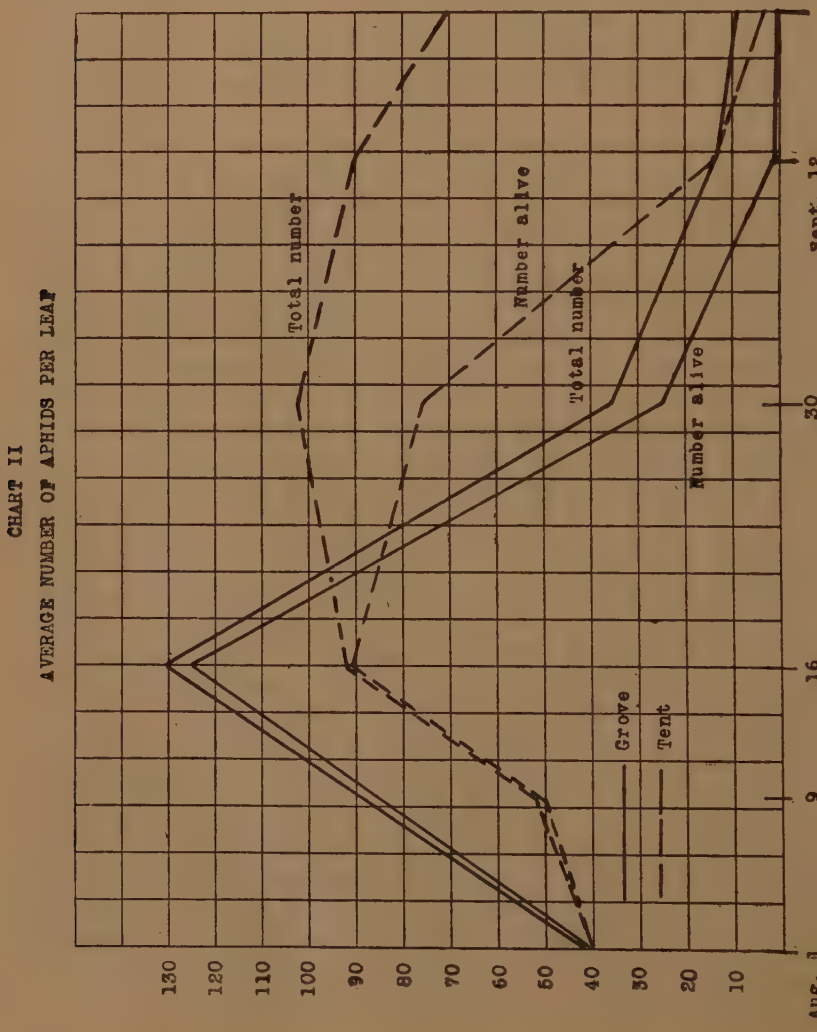
TABLE 4—WALNUT APHIS IN GROVE IN SANTA ANA, ORANGE COUNTY

Date	Number of leaves examined	Number of aphids per leaf	Percentage of aphids infected	Number of living aphids per leaf
August 1.....	60	41.35± 3.17	0.48±0.29	41.10± 3.17
August 16.....	10	131.70±16.86	4.56±1.07	125.90±16.86
August 30.....	20	35.25± 3.74	37.45±2.24	34.55± 2.68
September 12....	20	13.60± 2.21	93.38±1.60	0.90± 0.20
September 20....	10	8.80± 0.81	94.45±1.24	0.20± 0

TABLE 5—WALNUT APHIS IN TENT 2, SANTA ANA, ORANGE COUNTY.

Date	Number of leaves examined	Number of aphids per leaf	Percentage of aphids infected	Number of living aphids per leaf
August 1.....	20	40.00±11.28	0.	40.00±11.28
August 9.....	13	50.23± 9.55	0.15± 0.36	50.15± 9.56
August 16.....	10	91.80±10.50	1.20± 0.30	90.70±10.33
August 30.....	10	102.30± 4.45	25.71± 2.95	76.00± 4.50
September 12....	10	89.80± 9.95	84.97± 1.99	13.50± 2.19
September 20....	10	70.50± 7.91	95.60± 1.16	3.10± 0.98

On August 1 it was found that the grove there was an average of slightly over 41 aphids per leaf, of which about one half of 1 per cent



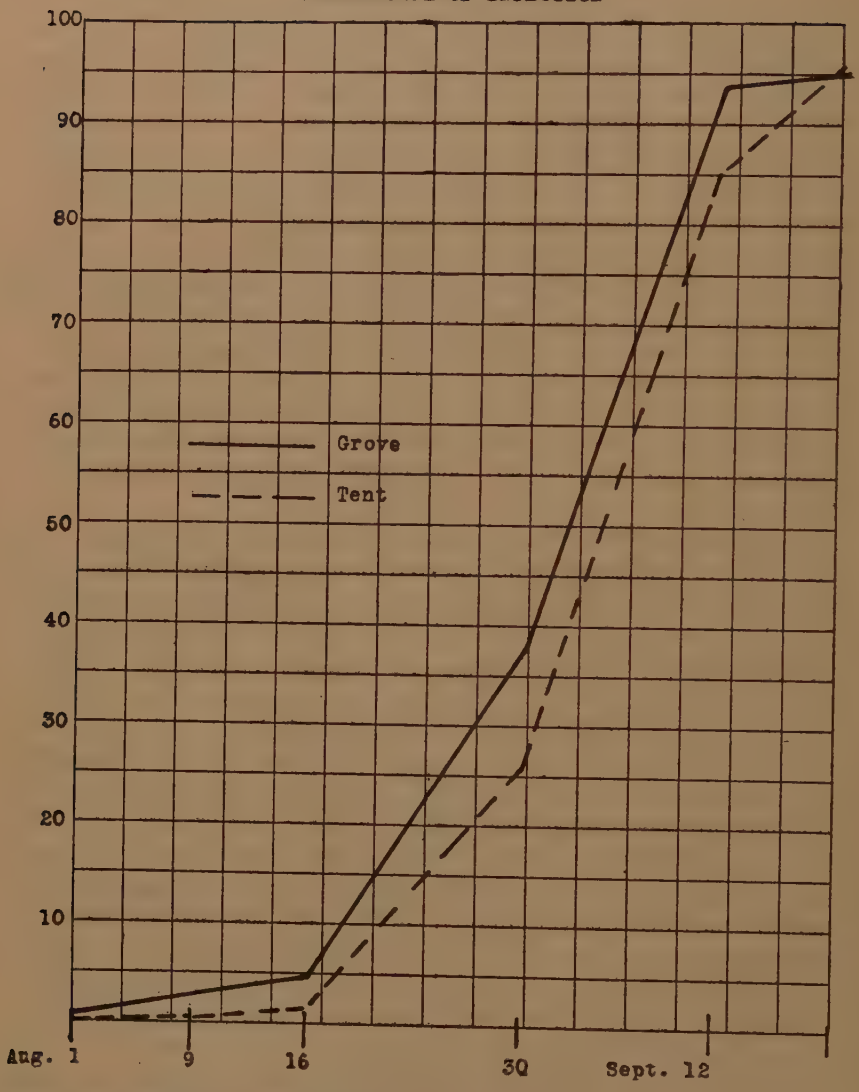
were infected with the fungus. In the following two weeks these increased greatly, until on August 16 there were present about 132 aphids per leaf. The percentage of infection had also increased, but not so rapidly, there being about 4½ per cent of the aphids killed by the fungus. In the next two weeks the number of aphids per leaf had dropped to but slightly over 35, of which some 37 per cent were infected. Two weeks later, on September 12, only about 13 aphids per leaf remained, and over 93 per cent of these were infected with the fungus, leaving an average of a little less than one aphid per leaf alive. By September 20 the number had increased to almost 9 per leaf, but again the percentage of infection was high, being almost 95 per cent, with the result that the aphids were practically eradicated.

This almost complete eradication is accounted for by the action of the fungus together with other natural agencies. Which of these was the most important the writers cannot state, but the presence of the mummified bodies of aphids infected by the fungus permits the interpretation that this parasite was largely responsible for this great mortality.

More convincing results are obtained from a study of Table 5. This is a record of counts made on the tree under tent 2, from August 1 to September 20. During observations covering a period of four months (June to September) no insect enemies of the walnut aphis were ever found in this tent. No specimens of *Coccinellidæ*, *Syrphidæ* or *Chrysopidæ* were seen, yet results parallel to those observed in the grove were found. On August 1 an average of 40 aphids per leaf was present and none were infected by the fungus. This tree had been sprayed in the spring before the buds opened with lime sulfur, with the result that no aphids appeared until very late. In fact it was at this time that the first real infestation was noticed. These probably were the succeeding generations of some few that gained entrance accidentally from time to time during the summer. By August 9 they had increased, as might be expected, to about 50 per leaf, with a small percentage infected with the fungus. A week later the number per leaf was found to be about 92, with about one dead as a result of fungus attack. Approximately 102 aphids per leaf were present on August 30, but by this time the percentage of infection had increased to 25.71 per cent, with the result that only 76 per leaf remained alive. These increased to about 90 aphids per leaf in the next two weeks, the number infected increasing as well, although in greater proportion. Almost 85 per cent were so infected, leaving but 13.5 per leaf alive. On September 20, when the last observation was made, there were found to be 70.5 aphids per leaf on an average, of which 95.6 per cent were infected by the fungus, leaving only slightly more than 3 aphids per leaf alive.

CHART III

PERCENTAGE OF INFECTION



Here the infestation had been almost eradicated without the aid of any predaceous insect enemies. Charts II and III show the amount of infestation and percentage of infection in the grove and in tent 2.

Other Hosts

Clinging to the under side of the walnut leaves was often found a small bark louse belonging to the genus *Psocus*. These bark lice

were examined from time to time as they were found, and without any exception they were infected with this same *Entomophthora*.

Description of the Fungus

During the spring of the year and in places where the humidity is rather high due to the fogs, an *Entomophthora* is parasitic upon the walnut aphis, *Chromaphis juglandicola* (Kalt.). The disappearance of this insect during the summer months was always thought to be due to heat and predaceous insects. Only one record has been found where a fungus was responsible for the control of this pest.[1] The writers have been unable to learn whether or not this was the same fungus as that herein described.

This fungus attacks the aphid in all its stages of growth from the mature winged form to the small first instar larva. One of the first signs of parasitism of the insect is the sluggish movement of the individual attacked. Later the body turns a darker yellow and is somewhat swollen. Then from the body of the dead aphid the small hyphæ begin to protrude, which in due time form a white fringe about the body (pl. 9, fig. 1). In some cases the protruding hyphæ are in such numbers that the insect looks like a white, glistening, spherical mass, which later turns a light brown in color. When the bodies of the insects are crushed and placed under the microscope, they are seen to be filled with large septate anastomosing hyphæ. These mummified bodies remain for some time, hanging to the leaves by means of haustoria until removed by some external agency.

The spores of the fungus are bell-shaped with a sharply pointed apex (pl. 9, fig. 2). These spores are borne on a club-shaped conidiophore. When the spore is mature, it is shot from the conidiophore with considerable force. Clinging to the spore is a mass of protoplasm that glues it to any object which it happens to strike. If the object happens to be the body of an insect, the spore germinates and the germ tube penetrates the insect. If, however, the spore happens to strike an unfavorable object for its growth, it again forms a secondary spore (pl. 9, fig. 3), which is much smaller but is ejected in the same manner, and the process is repeated until the spore either is exhausted or finds a suitable host. On the wings and legs of the affected insects could be seen many of the discharged spores imbedded in a cushion of protoplasm. Many of these spores had germinated and produced the secondary spores.

Some of the dead aphids were black and were thought to be attacked

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[1] Davidson, W. M. Walnut aphids in California. U. S. Dept. Agr., Bull. 100, page 35, 1914.

by a *Cladosporium*, but when examined under the microscope they were found to be filled with dark brown spherical bodies about 30 microns in diameter. These spherical bodies are believed to be the resting spores of the fungus. An attempt was made to germinate the resting spores but with no success.

Entomophthora chromaphidis n. sp.

Conidia bell-shaped with broad truncate base and sharply pointed apex; 11–14 microns by 10–11 microns; usually containing a single large oil globule, and surrounded after discharge by a mass of protoplasm. Conidiophores simple or compound; broad at the apex and gradually tapering to a narrow base; producing white or brownish masses which may or may not coalesce over the body of the host. Secondary conidia oval with rounded apex and formed by direct budding from the primary spore.

Resting spores, azygospores, brown, 30 microns in diameter. Host attacked by rhizoids.

Host: *Chromaphis juglandicola* (Kalt.), *Psocus* sp.

Habitat: California.

Explanations to Plate 9

Fig. 1. The fungus growing from the body of an aphid making a white fringe.

Fig. 2. A discharge spore embedded in the ejected mass of protoplasm.

Fig. 3. A discharge spore producing the secondary spore.

Summary

1. In the spring of 1917 the walnut groves of Southern California were heavily infested by *Chromaphis juglandicola* (Kalt.).

2. Although a period of extreme heat in June, and the presence of numerous insect enemies throughout the season were responsible for the death of a large percentage of these aphids, it was noted that a fungus also contributed to their mortality.

3. Before the period of extreme heat, in El Monte, as high as 88 per cent were killed by this fungus (*Entomophthora chromaphidis* n. sp.).

4. Some time after this period of extreme heat, the aphids increased rapidly but under the conditions noted were effectively controlled by this fungus.